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Color Discrimination in *Citellus lateralis chrysodeirus* (Merriam, 1890)

By John H. Wirtz

Eingang des Ms. 17. 4. 1967

Of all visual processes investigated, color vision has received the most attention. All sciurids, with the exception of the flying squirrel *(Glaucomys)* have pure cone retinae; yet there is little work reported on color vision in this group. Kolosvary (1934) found when the East European ground squirrel, *Citellus citellus*, was offered variously colored bits of paper for nesting material, certain colors were selected more readily than others. From these data he concluded that this form possessed color vision. Walls (1942), in comparing these data with color preference in the tree-squirrel, indicated some doubt as to the validity of Kolosvarys interpretation. Reimov (1957), working with the dwarf souslik *Citellus pygmaeus* in the field, indicated that this animal exhibited a differential brightness discrimination rather than a response to color.

Fig. 1. Citellus lateralis chrysodeirus (Merriam, 1890)

Because of the diurnal habit and the fact that golden mantled ground squirrels (Fig. 1) have photopic retinal mechanisms, it would be reasonable to suspect that they would demonstrate color vision. Arden and Tansley (1955) by means of electroretinograms worked out the spectral sensitivity of the pure-cone retina of the grey squirrel, *Sciurus carolinensis leucotis,* and found that there was good color sensitivity. And yet, when tests for responses to color are conducted, there is little if any indication that squirrels actually see color, at least as color is interpreted by man.

Color Discriminations Testing

Three approaches were used in an attempt to ascertain whether or not mantled ground squirrels would indicate a color preference. In the first attempt at color discrimination, ground squirrels were conditioned to coming to a particular area to obtain unshelled peanuts. After the conditioning period, unshelled peanuts stained blue, red, yellow, and green with Rit dyes were substituted for natural colored peanuts. Colored and uncolored peanuts were also soaked in gasoline, pine pitch, alcohol, formalin, creosote oil stain, and pine oil disinfectant, in order to eliminate the possibility of these animals responding to odor. In another experiment, colored and uncolored peanuts were placed in No. 11 veterinary capsules and suspended from strings. This eliminated not only odor, but also prevented the animals from "recognizing" peanuts by feel and/or texture.

In the third experimental set-up, three plywood boxes were constructed, each with a door through which ground squirrels could enter (Fig. 2). Once the animals were conditioned to go into the boxes for food, colored construction paper was placed over each door. At first, red, yellow, and blue colors were used. Later, other colors plus various shades of grey and finally no colors at all were employed. To test for color discrimination, only one box, the reward stimulus, had food in it which was available to the animal. The non-reward stimuli had food in the box, but it was not available

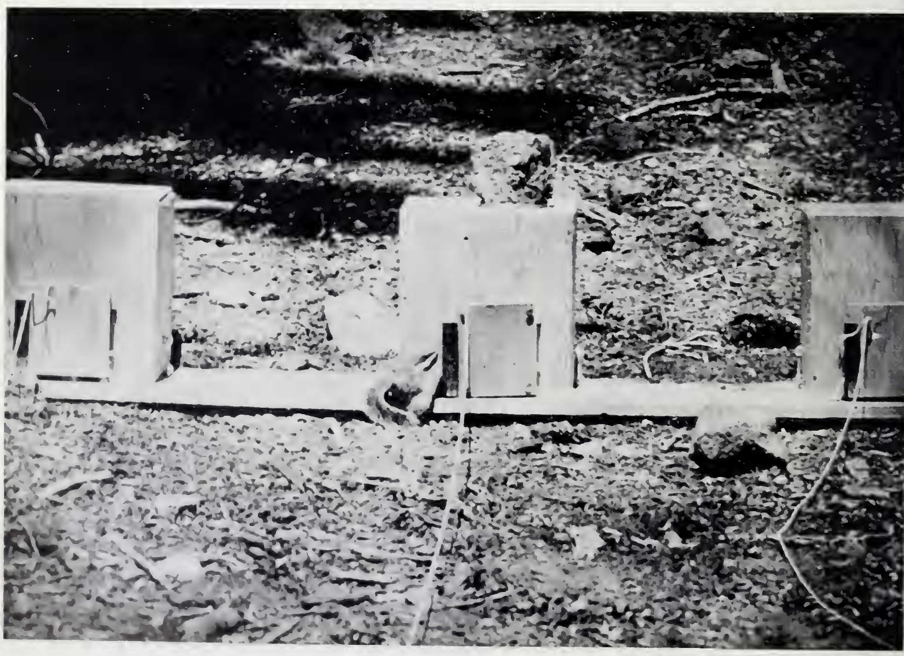

Fig. 2. An illustration of the experimental units and the manner in which they were arranged when testing for color vision

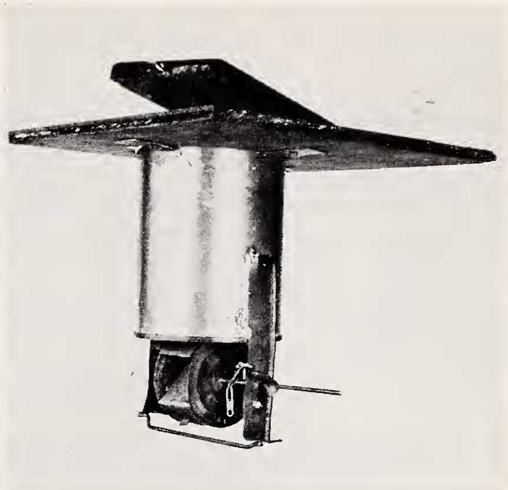

Fig. 3. Photograph of the food dispensing mechanism. Food was put into the container through an opening at the top

to the ground squirrels. After each trial, whether successful or not, the positions of the boxes were changed, thus avoiding the possibility of position-learning.

When the boxes were fiirst set out, generous amounts of the food, peanuts and oats mixed, were placed on the floor of the box. This soon proved to be impractical; for once an animal entered the box, it would stay there until its cheek pouches were full. To overcome this difficulty, a food dispensing device was invented (Fig. 3 and 4). This consisted of a tin can as a reservoir with a one inch opening cut into the bottom. Beneath this opening, a wheel two inches in diameter, and one inch wide, with a pocket in it large enough to hold five peanut kernels was suspended on an axle between brackets. A short lever was soldered to the axle, and a cord was attached to the inside of the door and the lever. Thus, when the door was opened, five peanuts were dropped to the floor of the box. When the device was first set up, the operation was entirely automatic, i. e., the bucket would trip on opening the door and be reset when the door closed. This proved to be disadvantageous, for the animals soon learned to respond to the sound of dropping peanuts as they opened the door to leave the box. They would then turn around and continue to fill their pouches. This problem was overcome by attaching a length of string to the lever ("A" Fig. 4) and by manually operating the closing of the door and resetting of the bucket.

Experimental Results and Discussion

I. Dyed peanuts

a. Training period: At the beginning of the color discrimination experiments, unshelled, non-colored peanuts were put on a board in an open area and left there until a number of animals were conditioned to coming to the area.

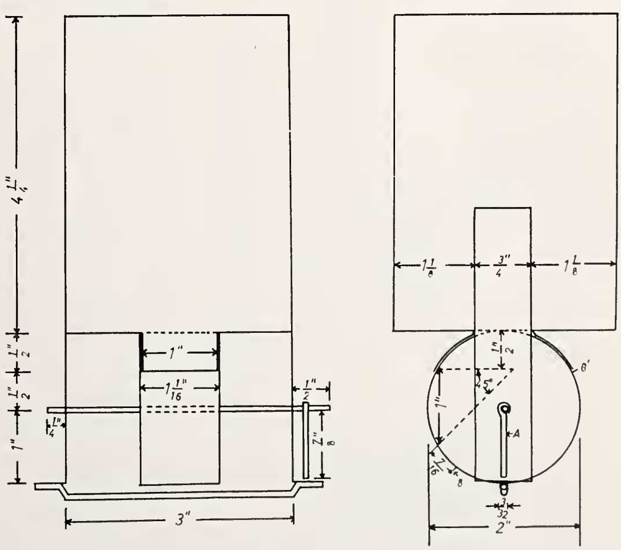

Fig. 4. Construction details of the food dispensing device used in color vision experiments

b. *Peanuts dyed with methyl green:* After the animals were assumed to be conditioned, the uncolored peanuts were exchanged for some which had been dyed with methyl green.

c. *Varicolored peanuts in individual piles:* After several animals habitually visited the area and were conditioned to green peanuts, peanuts dyed purple with gentian violet, red with methyl red, and black with India ink were also placed on the board. The variously colored peanuts were placed in individual piles to see if the animals showed any preference for the different colors. This technique did not work well, for the animals would come in from the same direction, and stop at the first group of peanuts, regardless of color.

d. *Varicolored peanuts in one pile:* The peanuts were then put in one pile and mixed. However, the animals still indicated no color preference, taking the first peanut they came to.

II. Deodorized peanuts

It was then suspected that odor was a more important factor than sight in detecting the food. To observe if this were the case, uncolored and colored peanuts were soaked in either gasoline, formalin, 95 percent alcohol, or rubbed with pine pitch. The assumption was that these odoriferous substances would mask the scent of the peanuts, and if color perception were present, only green peanuts would be taken. The results were entirely unexpected. The treated peanuts were taken just as readily as untreated peanuts.

III. Discrimination boxes

Because the animals display a considerable manipulative ability, it was decided that if there were some device in which doors must be opened, and if on these doors there were colored cards, perhaps the animals would demonstrate color discrimination by consistently choosing the box with the colored door to which they had been conditioned, and thus be rewarded with food.

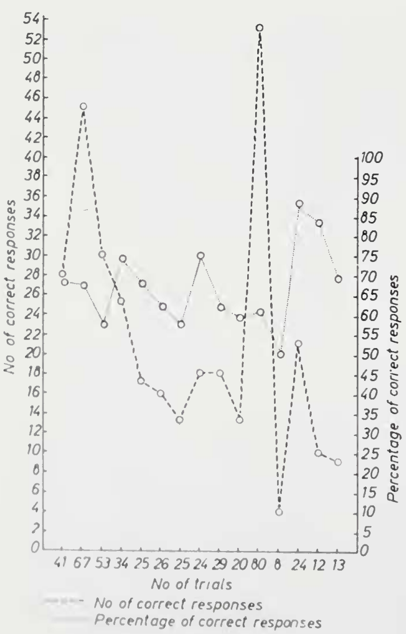

Fig. 5. A graph showing the number of correct responses to the experimental unit, and the percentages of right choices

Three boxes, nine inches on a side, were constructed. At first food was just placed inside the experimental box. This proved inefficient since the amimals would sit inside and fill their cheek pouches. It was then that the food dispensing devise was constructed. Figure 5 is a graph showing the number of right responses to the experimental box, as well as the percentage of correct choices. The graph was plotted for the total of the 481 trials conducted over a 15 day period. There were 320 correct responses and 161 wrong choices. Of the wrong responses, 78 were made to the right of the experimental unit, and 81 to the left of it. The average percentage for all of the trials is 66.5 percent.

a. *Training period:* On the first day of the trials, the boxes were set out loaded with peanuts and the doors tied open, thus getting the animals conditioned to going into the boxes for food. Three hours later the doors were closed, but each box still had food in it. At first the ground squirrels investigated the area around the base of each box until they learned to open the door. Once they learned to open the doors, the animals were conditioned to a blue color.

b. *Blue card:* With a blue card on the reward stimulus and no colored cards on the non-reward stimuli, there were 11 correct responses and four wrong ones.

c. *Blue versus red and yellow cards:* Next, red and yellow cards were placed on the non-reward stimuli. For the six days that this combination of colors was used the number of correct responses was greater than wrong choices, of the 227 trials run, 150 were to blue, 25 to red and 52 to yellow (Table 1). To avoid the possibility of position learing, the boxes were shifted at random after each trial.

d. *Blue cards on all boxes:* After the sixth day and for the next three days, blue cards were substituted for red and yellow cards on the non-reward stimuli. The first day's results netted 13 right and 13 wrong responses. However, on the second day, there were 18 right and six wrong choices, indicating that the animals were respon-

The Number of Responses to Colored Cards on the Experimental Unit

Day	No Colors	Blue* (only)	Blue* Red Yellow	Yellow* (only)	Green* (only)	Pink (only)	Gray (only)	All Gray	Green* Gray Pink	Pink* Gray Green	Gray* Green Pink
1		11 r 4 w	17 r 9 w								
2			45 r 19 w								
3			30 r 21 w								
4			25 r 10 w								
5			17 r 8 w								
6			16 r 10 w								
7		13 r 13 w									
8		18 r 6 w									
9		14 r 12 w		4 r 2 w							
10				1 r 0 w	2 r 0 w	2 r 1 r	6 r 6 w	3 r 6 r			
11	11 r 7 w						28 r 17 w	11 r 4 w	7 r 2 w		
12									4 r 4 w		
13									7 r 3 w	8 r 0 w	6 r 0 w
14	10 r 2 w										
15					9 r 4 w						

The colors across the top of the chart are those used on the doors of the boxes. The asterisk after the color indicates the reward stimulus. The other two colors were on the non-reward stimuli. The word "only" beneath the color indicates there were no colors on the controls. The letter "r" after the numbers in the columns indicates the number of correct responses; the letter "w", the number of wrong responses.

ding to some stimulus other than color. Results of the third day were similar to those of the first with 14 right and 12 wrong responses.

e. *Yellow card:* Next, a yellow card was substituted for the blue card on the reward stimulus. Immediately after the change of cards, two correct responses were made to the yellow.

f. *Pink card:* A pink card was exchanged for the yellow one following the responses to the yellow card. There were two correct choices and one wrong response.

g. *Gray card:* After exchanging the pink card for a gray one, there were five right and six wrong responses.

h. *All gray:* After these 11 trials, gray cards were also placed on the non-reward stimuli. Of the nine trials run, three were correct and six were wrong.

i. *Gray card only:* The following day the gray cards were removed from the non-reward stimuli. Of the 45 trials conducted, 28 responses were correct and 17 were wrong.

j. *All gray:* The gray cards were then replaced on the non-reward stimuli. The results from the 15 trials were 11 right and four wrong.

k. *No color:* After these 15 trials, the cards were removed from all units. Following this there were 11 right and seven wrong choices.

l. *Green versus gray and pink:* Following the trials with no colored cards on the units, a green card was placed on the reward stimulus, and gray and pink cards on the non-reward stimuli. Of the 27 trials run over a three day period, there were 18 right choices and nine wrong ones.

m. *Pink versus gray and green:* With a pink card on the experimental unit, there were eight correct responses and no wrong choices.

o. *No colors:* Again after removing all cards, there were 10 right responses and two wrong choices.

p. *Green card only:* Finally, a green card was put on the reward stimulus and no cards on the other two units. The results obtained were nine right and four wrong responses.

The data of this experiment were subjected to the chi-square test, and at the five percent level with one degree of freedom, the results were significant. The significance cannot be interpreted to mean golden-mantled ground squirrels have color discrimination. It does mean, however, that some stimulus other than color vision was responsible for the animals making the right choices to the experimental unit.

Like many other animals, mantled ground squirrels have anal scent glands, and thus, mark an area for identification. Several attempts were made to destroy any odor the boxes might have had from animals being in contact with them. The boxes were "painted" with pine oil disinfectant, and later sprayed with air freshener (the type used in homes). In either case there was no change noted in the facility with which the animals found the experimental unit. Finally, the boxes were painted with creosote oil stain. At first, the animals, except number six, appeared confused. Number six went immediately to the experimental unit, but the others sniffed around the three boxes and did not open the doors. After number six returned to the experimental unit several times, other animals began to enter the box and help themselves to food.

Observations of the activiy of the animals would tend to indicate that smell is important. In general, there was considerable investigative behavior by all participants. Even after becoming conditioned to finding food inside the box, the animals would sniff around the base of the boxes before opening the door. At times, individuals would go directly to one non-rewarding unit and then to the other, back to the first unit, run away a short distance, stop and rest, and then proceed to the reward stimulus.

Animal number six, the dominant, behaved differently than the other mantled ground squirrels. He soon learned to operate the tripping mechanism of the dispensing device. When the animal opened the door of the reward stimulus, the vendor would spill a certain quantity of food on the floor, number six would begin to fill his cheek pouches, if there was room for more he would pull on the string which tripped the dispensing bucket, pick up the food dispensed, and continue this until his pouches were full; then and only then would he leave.

Oats were also tried as a food item, thinking that there was less odor and therefore not as readily detectable, however, there was no difference in the results.

IV. Peanuts on strings

In another series of experiments, animals were conditioned to reaching for peanuts suspended from strings. After the initial training, peanuts colored blue with Rit dye were used to replace the natural colored peanuts. In either case all peanuts were taken readily. After having been subjected to taking blue peanuts, some peanuts colored red and others yellow, also colored with Rit dyes, were likewise suspended from strings. At first the peanuts were all suspended at the same level above the ground. Later some red, some yellow and some of the blue peanuts were suspended high enough so that the animals had to "work" to get them. The nuts closest to the ground were taken first, and then those that were hung slightly out of reach. In either situation no color preference was indicated.

V. Peanuts in gelatin capsules

The final series of experiments were somewhat similar to those just described. The animals were again given blue colored peanuts. At the end of the training period, peanuts colored blue, red, or yellow and non-colored nuts were put into number 11 gelatin veterinary capsules, being careful to leave no peanut aroma on them. Some of the enclosed peanuts were then suspended above the ground within easy reach of the animals. Other capsules were placed in a pile on the ground.

The animals approached the suspended peanuts, sniffed briefly, and then either scurried off or began investigating the area. Similarly, the capsules were sniffed, and then abandoned. Only one capsule was taken and opened up, and it contained a yellow peanut. Some of the capsules were later smeared with peanut butter. Immediately animals tried to pick these up and carry them off, but because of the smoothness and size of the capsule this was impossible. After several attempts to carry the capsule away, the animals paid no further attention to them. These observations seem to indicate that smell, rather than color, is more important in finding food.

Perhaps reactions to color depend upon certain releasing mechanisms which are present in nature, but are not present in experimental situations. Tinbergen (1951), working with the butterfly, *Pieris brassicae*, showed that for oviposition the female selects green objects, but for feeding selects yellow, blue, and red flowers. Certainly, one cannot readily compare reactive behavior between arthropods and vertebrates. However, in the light of such findings, it may well be that if the experimental approach were changed, and color perception tests were conducted under more natural conditions, one might obtain entirely different results.

Conclusions

Although there were a statistically significant number of correct responses to the reward stimulus, it cannot be concluded positively that there is a color discriminatory capacity in golden-mantled ground squirrels. On the other hand, there is no proof that color vision is entirely lacking in these animals. LOCHER (1933) concluded that there is a very weak capacity for discriminating color in *Sciurus vulgaris*, but so very weak that, within limits of normal individual variation, it may be lacking in a particular individual. It may be that this is the case in mantled ground squirrels.

Suggestions as to methodology

One factor that appears to be responsible for the high number of correct responses is that the animals approached the experimental site from the same direction. Even though the boxes were shifted at random, over a long period of time the number of correct choices would be greater than wrong ones through choice alone. Suppose that at one trial the order would be: non-reward, non-reward, reward stimulus; the animal coming from the left could make the two wrong choices before a right response. On the other hand the animal coming from the right would make no wrong choices. If the position were changed to non-reward, reward, non-reward stimulus, both animals would each make a wrong choice and then a right one. Also, there would be a number of chance responses to the reward stimulus. Thus in no case are the animals responding to color but rather are "finding" the reward stimulus through the normal course of their behavior. A more satisfactory arrangement would be to control the direction of approach to the experimental units. This could be accomplished by elevating the boxes, and have runways, originating from a common point, leading to each of the units.

Odor appeared to be an important factor in finding the reward stimulus. Although attempts were made to mask the smell of peanuts, there was no assurance that this was done. A more satisfactory method of eliminating smell as a factor, would be to destroy the sensory structures of olfaction.

There is also the problem of color versus brightness discrimination. Prior to further field work it is suggested that all color cards be carefully matched for brightness value. In addition to color cards matched for brightness, a series of gray cards, not only matched with the color cards for brightness, but also an intermediate series should be used. The use of gray cards is a convenient method for eliminating the possibility of brightness discrimination. If the animal is trained to a color versus medium gray, and then other grays are substituted from a finely-graded series, color is qualitatively seen if no confusion arises between these grays and the positive stimulus.

Perhaps the best way to test for color vision would be to bring the animals into the laboratory, and use monochromatic light, the intensities of which can be carefully regulated.

The experimental units can also be used to test for pattern and form discrimination, and success or failure, might serve as a check on the validity of color experiments.

Summary

Although there were more correct responses during the experiments on color discrimination there is little evidence that golden mantled ground squirrels actually respond to a color stimulus. It appears that odor is a more important factor in making correct choices. The positive stimulus in some way, perhaps by glandular secretions from the fore paws, is marked, thus accounting for the high percentage of correct choices.

Zusammenfassung

Obwohl mehr richtige als falsche Diskriminationslösungen auftraten, wurde kein Beweis dafür gefunden, daß *Citellus lateralis* tatsächlich auf Farbreize reagierte. Es scheint, als ob der Ge-

ruch als Haupterkennungsfaktor diente. Die richtigen Lösungen erfolgen wahrscheinlich durch das Deponieren positiver Reize in Form von Drüsensekretionen der Vorderpfoten.

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Observations on the internal Structure of the Aorta in East African Mammals, with particular Reference to the Klipspringer, Gerenuk and Hippopotamus

By SYLVIA K. SIKES

Eingang des Ms. 11. 5. 1967

Introduction

During a recent field survey of cardiovascular disease in free-living wild mammals and birds in East Africa[1] a technique was developed for the comparative estimation of lipoidal and calcific deposits in the arterial wall in mammals. As a result of the consistent use of this technique throughout the survey, some peculiarities of structure of the aortae of certain species were noted.

The most outstanding of these were found in the klipspringer *(Oreotragus oreotragus* Neumann), the gerenuk *(Litocranius walleri* Brooke), and the hippopotamus *(Hippopotamus amphibius* Heller). It is possible that these specialisations of structure may have a functional relationship to the habits of the species concerned.

Materials and Methods

1. Collection of specimens

Specimens of free-living wild mammals were selected and shot in East Africa by the author, with the specific purpose of examining the cardiovascular system. To ensure minimal damage to the circulatory system, they were usually killed by means

[1] Financed by the British Heart Foundation, and carried out under the auspices of the Zoological Society of London.